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Plot size for progeny selection in maize (*Zea mays* L.)

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Summary. Six progeny trials that included 147 half-sib progenies of maize (*Zea mays* L.) population ESALQ PB-5 were conducted for the purpose of studying plot size and its consequences in recurrent selection programs. The progenies were evaluated in three 7 × 7 duplicate simple lattice experiments using one-row plots of 5 m^2. At harvest each plot was partitioned into five sub-plots (sampling units), and data was collected from each sampling unit. At the same time and place the same progenies were evaluated in three 7 × 7 duplicate simple lattice experiments using 1-m^2 (linear row with 5 plants) plots. Data were collected for plant and ear height, ear diameter, total ear weight, and total grain yield. The data were combined by using adjacent sampling units, and the analyses were performed by considered five plot sizes in addition to those of the independent trials with 1-m^2 plots. The experiments with 1-m^2 plots were less efficient in discriminating for yield traits among progenies than those with 5-m^2 plots. The combination of plot size and number of progenies evaluated indicated that an optimum plot size for yield was between 3 and 4 m^2, or 15–20 plants per plot. With such sizes the expected gain was maximized for the four replications used in this study. If the total area covered by each progeny is constant, the maximum gain from selection, however, is attained by decreasing plot size and increasing the number of replications. The minimum size of plots is, however, limited by practical or theoretical criteria. Plot size affected the estimates of additive genetic variance, coefficient of heritability, and genetic coefficient of variation for all of the traits. No practical limitation was observed for conducting experiments with 1-m^2 plot.

Key words: Plot size – Progeny selection – Genetic parameters

Introduction

The statistical and experimental techniques applied to plant breeding have contributed greatly to the development and accuracy of selection methods. Plot size is an important component of selection programs. It depends on factors such as the nature of the treatments, the availability of experimental area and resources, and statistical precision required for testing hypotheses and estimations.

Several authors have shown an inverse relationship between plot size and experimental error (Day 1920; Smith 1983; Steel and Torrie 1960; Hatheway 1961; Le Clerg et al. 1962; Le Clerg 1967), but the asymptotic nature of such a relationship indicates that little is gained in precision by increasing plots that are sufficiently large. Federer (1955) considered the main factors in the determination of plot size to be the nature of the experimental material, the variability of individuals within plots in comparison to variability among plots, and the cost per individual relative to the cost per experimental unit. Other factors of a practical order such as the manner of conducting the experiments must also be considered. In progeny evaluation trials an important factor limiting plot size is the number of seeds available per progeny, which may dictate the use of smaller plots and/or fewer number of replications.

There are several methodologies available for determinating the adequate size of experimental plots. The method of Smith (1938) is based on the relationship between the variance of plot means and plot size. Thus,

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taking V as the variance of the smaller units in a uniformity trial, the variance of plot means, with plots comprising × adjacent units, is given by $V_{\bar{x}} = V/x^B$, where B is an index that measures soil variability and varies from 0 to 1. From an estimated B, the optimum plot size is given by $x = Bk_1/(1-B)k_2$, where k_1 and k_2 are factors of proportional cost related to the number of plots per treatment and total area per treatment, respectively.

Federer (1955) showed how to determine plot size through the methodology of maximum curvature, which is based on the relation between the coefficient of variation (CVx) and plot size (x) for plots of varying sizes in a uniformity trial. Other methods for determining plot sizes have been provided by Keller (1949), Hatheway (1961), Lessman and Atkins (1963), Meier and Lessman (1971), Pimentel-Gomes (1984), and Silva et al. (1984). Koch and Rigney (1951) showed how to estimate the soil heterogenity coefficient (Smith 1983) from experimental data when treatments are in split-plot and lattice arrangements.

Some reports dealing with plot size determination in maize (*Zea mays* L.) have presented results that were to a certain, extent consistent. Hallauer (1964) collected data from 217 experiments at 15 locations for 4 years in Iowa and used the estimates of the coefficient of variation and soil variability (Smith 1938) to find convenient plot size. Eberhart (1970, 1971) showed the asymptomatic pattern of the expected gain from selection on plot size variation and that littel is gained with more than 15–20 plants per plot. Marquez-Sanchez (1972) found the optimum size for obtaining parameter estimates with acceptable precision to be around 17 plants per plot. Días (1978) found that the optimum plot size for single crosses, double crosses, varieties, and composites of maize was similar, with some variation among traits; approximately 13, 15, and 5 plants per plot were needed for 50-kernel weight, total yield (ear weight), and plant height, respectively. Storck and Uitdewilligen (1980) used several methods and determined that a plot size of 5 m^2 was needed for yield. For the intercropping of maize and common beans Zimmermann (1982) found 18 m^2 to be the optimum plot size for maize yield. Ribeiro et al. (1984) found 3.5 and 4 m^2 to be the appropriate size for maize in single crop and intercropped with cowpea. In experiments comparing the genetic material of maize in Brazil plots not larger than 10 m^2 have generally been used, and progeny evaluation plots of 4–5 m^2 (20–25 plants per plot) have been used without any compromise of the precision of experiments.

The use of reduced plot sizes in plant breeding programs is sometimes imposed by the number of entries in experimental trials. The development of experimental techniques, the use of computer analysis, and the expansion of breeding programs have led to an increasing trend in the number of experimental entries; in maize these are represented either by progenies in recurrent selection or by inbred lines and crosses in a hybrid breeding program. Under the hypothesis that smaller plots would allow the evaluation of a larger sample of genotypes, the present work was conducted with the object of analyzing the consequences of larger sample sizes and determining the appropriate plot size for selection.

Material and methods

These base population was ESALQ PB-5, a composite formed by intercrossing three short plant varieties ('Piracar-I', 'Eto Blanco' and 'Mezcla Amarilla'). A sample of 147 half-sib families was partitioned for evaluation in three duplicated simple lattice (7 × 7) experiments at one location. Each plot was represented by a linear row 5 m long with 1.0 m between rows; there were 25 plants per plot after thinning. Three experiments with the same progenies and design were conducted with linear rows of 1 m^2 with 5 plants after thinning. The experiments were indentified as 51, 52, and 53 for 5- m^2 plots and as 11, 12, and 13 for 1-m^2 plots, where the first digit indicate the plot size.

In experiments 51, 52, and 53 each plot was partitioned into five sample units of 1 m^2 each. In experiments 11, 12, and 13, the experimental unit was the plot itself. Data were taken for the following traits: ST, final stand; EN, ear number; EW and GW, total yield as measured by ear weight and grain weight, respectively, on a sample unit basis; ED, ear diameter, averaged on an individual plant basis; PH and EH, plant and ear height, respectively, where only one plant per sample unit was taken in experiments 51, 52, and 53 and averaged on an individual plant basis, and all plants were taken in experiments 11, 12, and 13.

The analyses of variance were performed according to the randomized complete block design, because the lattice design did not show efficiency in relation to the randomized complete-block design. For EN, EW and GW, the statistical analyses were performed after the correction of observations for stand variation (correction to ideal stand) following the covariance technique (Steel and Torrie 1960). Each experiment with plots of 5 m^2 gave rise to five analyses, corresponding to plot sizes varying from 1 to 5 m^2. For PH and EH, only the analyses for 5 m^2 and 1 m^2 were considered. The independent analyses representing three samples of 49 progenies were pooled in a grouped analysis after the observations had been adjusted for the general mean of the three experiments; the adjustment procedure was of the multiplicative nature, as suggesting by Chaves (1992) and Chaves et al. (1989).

For each grouped analysis of variance the following parameters were estimated: general mean (m); *F* statistic; experimental coefficient of variation (CV_e); standard error of treatment means (σ_m); genetic coefficient fo variation (CV_g); additive genetic variance (σ_A^2) and its standard error, $s(\hat{\sigma}_A^2)$; coefficient of heritability on progeny mean basis (h_m^2) and its standard error, $s(\hat{h}_m^2)$; and expected gain from selection in percent of the mean (g%) for a selection differential equal to one phenotypic standard deviation. Estimates of means and variances were given on a per plant basis.

The estimated parameters were used to compare experiments with different plot sizes for two different methods. The first method compared independent experiments by testing the difference in the discrimination power through the Schumann-Bradley test; this test is based on the estimated parameter $\hat{w} = F/F'$ that is compared with tabled values for significance, where *F* and *F'* are values of the *F* statistic obtained from similar experiments (Schumann and Bradley 1959). For the second method, estimates of the parameters were used to determine the trend variation for varying plot sizes. In these cases only a combination of adjacent sample units from experiments 51, 52, and 53 were used.

An optimum plot size for recurrent selection programs is proposed that is based on the maximization of the expected gain from selection as function of plot size and the proportion of selected progenies. The parameter g% was correlated to plot size through the function $g\% = a'x^{b'}$, where x is the plot size in m^2 and a' and b' are parameters of the equation. The expected gain from selection in percent of the mean is then given by $gs\% = i_p\ g\%$ or $gs\% = i_p\ a'x^{b'}$, where i_p is the selection differential in standard units and is a function of the proportion (p) of selected progenies (Falconer 1972). The most adequate plot size is determined through the combination of values of p and x that maximizes the expected gain from selection for a given situation.

The results obtained by the proposed method were compared with those obtained by the algebraic method of maximum curvature of the CV function, as suggested by Meier and Lessman (1971). The experimental coefficient of variation was related to plot size through the function $CV = ax^b$, and the point of maximum curvature was obtained by $x_c = a^2b^2(2b-1)/(b-2)]^{1/(2-2b)}$. It can be shown that by keeping the means of experiments with different plot sizes constant the parameter b is equal to in absolute value to half of the soil heterogeneity index of Smith (1938), or $b = -B/2$. Because B varies from 0 to 1, the range for b is -0.5 to 0.

Results and discussion

Plot size and the discrimination power of the experiments

Table 1 shows the estimated parameters ($\hat{w}$) for the Schumann-Bradley (1959) test that were obtained from the combined analysis of variance by comparing experiments of varying plot sizes in relation to the independent experiments of 1-m^2 plots.

For plant height (PH) and ear height (EH), no significant differences were observed between experiments with 1-m^2 and 5-m^2 plots; such results were expected because the number of plants for those traits were approximately the same in both experiments. For other traits the $\hat{w}$ estimates showed an increasing trend with increasing plot size, indicating a higher power for discrimination among treatments for plots of larger sizes. Such a trend is due to a reduction in the error variance, achived by increasing plot size, which leads to higher *F* values in the experiments with larger plots. Nevertheless, significant differences for the discrimination power only were observed for ear diameter (ED) and grain yield (GW) for plots equal to or larger than 3 m^2 and for ear weight (EW) for plots with 5 m^2.

The significance limits for $\hat{w}$ in the tables given by Schumann and Bradley (1959) have a maximum real number for the error with 40 degrees of freedom. The *F* values used to estimate w were obtained from 429 (grain yield and ear weight) and 432 (ear diameter) degrees of freedom for the error. Therefore, we used infinite as the number of degrees of freedom. Our results always took a 1-m^2 plot as the base for comparison. The power to discriminate between experiments with plot sizes between 1 m^2 and 5 m^2 when 5 m^2 was taken as the base for comparison was not possible because varying plot sizes from independent experiments were not available. Such a comparison would, however, be of interest to determine the limit of reduction in plot size without compromising the discrimination power of the experiment.

Table 1. Estimates of w for the Shumann-Bradley test obtained from pooled experiments of 5-m^2 compared to 1-m^2 plots

Traits	Pooled experiments				
	5 (1)	5 (2)	5 (3)	5 (4)	5 (5)
PH[a]	–	–	–	–	1.131
EH[a]	–	–	–	–	0.910
EN	1.091	1.212	1.354	1.267	1.319
ED	0.792	1.300	1.732**	1.999**	2.312**
EW	1.052	1.055	1.255	1.310	1.834**
GW	1.133	1.360	1.467*	1.611**	2.051**

*,** Significance levels of 5% and 1% respectively for infinite number of degrees of freedom for error

[a] Data from 5 plants per plot

(1), (2), . . . , (5): number of sampling units

Plot size and trend of the estimates of parameters

Estimates of statistic-genetic parameters for ear diameter (ED) and grain yield (GW) were obtained from pooled mean squares in the analysis of variance for each plot size (Table 2). The degrees of freedom were 144 for progenies within experiments and 429 (GW) and 432 (ED) for the pooled error variance.

The *F* statistic (variance ratio) indicates directly the power of the experiment for discriminating among treatment means. It showed a consistently increasing trend for both traits with increasing plot size. The *F* estimates were always greater for ED than for GW, and this difference also showed in increasing trend. The increasing trend for *F* is due to the decrease in the error variance with increasing plot size; an inverse relationship that has been reported by others (Day 1920; Smith 1938; Hatheway 1961; Le Clerg et al. 1962; Le Clerg 1967). Because ED is less affected by environmental effects, the decrease in the error variance for ED was greater than for GW, thus resulting in larger differences between the *F* estimates (between traits) for increasing plot sizes.

The standard error (σ_m) of the treatment means is a powerful indicator of experimental precision (Steel and Torrie 1960) because it takes into account the number of replications. The coefficient of variation (CV) also is a measure of experimental precision, but the number of replications is not considered in the CV. Because all of the comparisons between experiments included for replications, both σ_m and CV showed a similar pattern of variation: an asymptotic and decreasing trend with increas-

Table 2. Estimates of statistical-genetic parameters for ear diameter and grain yield obtained from pooled analysis of varying plot sizes

Estimates	Ear diameter[b]					Grain yield[c]				
	(1)	(2)	(3)	(4)	(5)	(1)	(2)	(3)	(4)	(5)
F	1.620	2.660	3.544	4.091	4.730	1.292	1.552	1.667	1.838	2.339
$\hat{\sigma}_m$	0.134	0.094	0.079	0.070	0.064	11.46	8.860	7.718	7.109	6.224
CV%	6.126	4.296	3.647	3.214	2.935	19.82	15.37	13.65	12.69	11.28
CVg%	2.413	2.768	2.909	2.826	2.834	5.358	5.708	5.613	5.797	6.527
$\hat{\sigma}_A^2$	0.445[a]	0.581[a]	0.641[a]	0.605[a]	0.609[a]	153.5	173.2	161.2	169.3	212.2
$s(\hat{\sigma}_A^2)$	0.145[a]	0.112[a]	0.106[a]	0.095[a]	0.091[a]	87.09	60.90	49.49	45.61	44.70
h_m^2%	38.28	62.41	71.79	75.55	78.86	22.63	35.55	40.35	45.58	57.25
$s(h_m^2\%)$	8.355	5.089	3.820	3.310	2.862	10.48	8.728	8.077	7.370	5.789
g%	1.493	2.186	2.465	2.456	2.517	2.550	3.403	3.566	3.913	4.939
m	4.472	4.354	4.351	4.351	4.353	115.6	115.3	113.1	112.2	111.6

[a] Values multiplied by 10
[b,c] Mean and variances in cm and g/plant, respectively
(1), (2), ..., (5): number of sampling units

ing plot size. The inverse relationship between CV and plot size is well known for several traits in different species (Federer 1955; Lessman and Atkins 1963; Meier and Lessman 1971; Storck 1979).

The genetic coefficient of variation (CV_g) is estimated by $\hat{\sigma}_g/m$, where σ_g is the square root of the genetic variance among progeny means; for half-sib progenies and negligible epistasis $\hat{\sigma}_m^2$ estimates one-fourth of the additive genetic variance ($\hat{\sigma}_A^2$). Since the means (plant basis) for both traits were nearly constant among experiments (Table 2), CV_g and $\hat{\sigma}_A^2$ showed a similar pattern of variation. This variation was random because theoretically the additive genetic variance is not expected to vary among environments except for scale effect; no apparent relationship was observed between CV_g or $\hat{\sigma}_A^2$ and plot sizes. The standard error of the additive genetic variance, $s(\hat{\sigma}_A^2)$, however, showed a consistently decreasing trend for ED and GW with increasing plot size. Therefore, the precision of the estimates of both $\hat{\sigma}_A^2$ and CV was improved for larger plots because of the smaller mean squares for both treatments and error, since the number of replications and degrees of freedom were the same for different analyses. Marquez-Sanchez (1972) reported that with six replications the number of plants per plot should be 17 to obtain an acceptable precision of the estimates of genetic parameters. Similarly, for values of $s(\hat{\sigma}_A^2)$ for plots larger than 3 m^2 (15 plants), the gain in the precision of the estimates was relatively low (Table 2).

The coefficient of heritability on a progeny mean basis (h_m^2) expresses directly the relative genetic variation of the selection units when selection is based on progeny means. Estimates of h_m^2 showed an increasing trend for both traits with increasing plot size, and ED always showed a higher heritability than GW for a given plot size. On the other hand, the standard deviation of the estimates showed a decreasing trend, although the decrease was very low for plots larger than 3 m^2, which approximates the size reported by Marquez-Sanchez (1972) for an acceptable precision of the estimates.

A close relationship existed between the pattern of variation of h_m^2 and F (Table 2). There was a mathematic equivalence between them, that is $h_m^2 = 1 - F^{-1}$, or $F = (1 - h_m^2)^{-1}$. Wearden (1960) also showed a mathematical relationship between F and h^2 (coefficient of heritability on individual basis).

The expected gain from selection (g) for a selection differential equal to one phenotipic standard deviation is estimated by $\hat{g} = h_m^2 \hat{\sigma}_F$. These estimates showed an increasing and asymptotic trend with increasing plot size, and in percent of the mean (g%) they were always higher for GW than for ED, despite the higher heritability of ED. The smaller expected gain for ED was due to the lower phenotypic variability and indicates that the coefficient of heritability itself is not always a powerful indicator of the expected results of selection.

The observed variations in the estimates of all of the parameters considered agree with the theoretical expectation. Methodologies of plot size determination have considered only parameters of statistical nature, such as the coefficient of variation and the error variance. Information relating the pattern of variation of genetic parameters estimated on an experimental basis with varying plot sizes was not found in the literature.

Plot size determination

The relationship between CV and plot size for ED and GW are shown in Figs. 1 and 2, respectively. The determination of plot size following the methodology given by Meier and Lessman (1971) led to values of 1.84 m^2 for ED and 3.64 m^2 for GW, indicated, by algebraic determination, by the points of maximum curvature of the

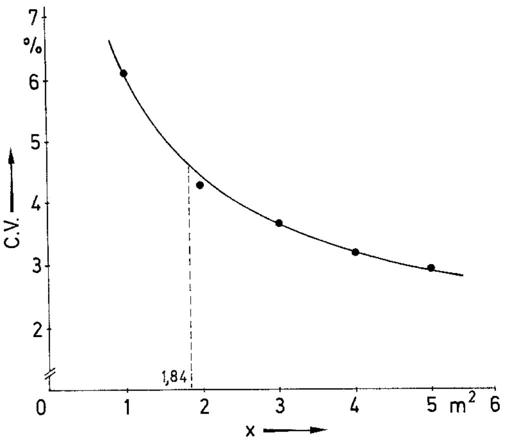

Fig. 1. Relationship between the coefficient of variation (C.V.) and plot size (X) for ear diameter (ED). —— Linie estimated by the function C.V. $= 6.04\ X^{-0.4554}$; o Observed points ($r^2 = 0.9971$ **); Point of maximum curvature: $X_c = 1{,}84\ m^2$

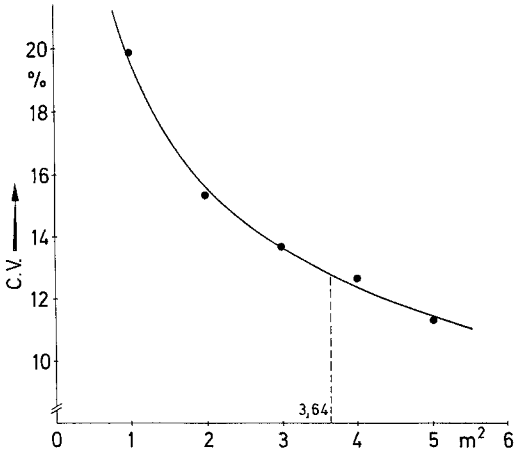

Fig. 2. Relationship between the coefficient of variation (C.V.) and plot size (X) for grain weight (GW). —— Line estimated by the function C.V. $= 19.72\ X^{-0.3371}$; o Observed points ($r^2 = 0.9938$ **); Point of maximum curvature: $X_c = 3.64\ m^2$

functions $CV = 6.04\ x^{-0.4554}$ and $CV = 19.72\ x^{-0.3371}$, respectively. The highly significant values for the coefficient of determination (r^2) of the regression functions showed that few points, as in the present case, are enough for a chose adjustment of the CV equations as functions of plot sizes.

The coefficient of variation as the basis for determining plot size has been properly used for comparisons among treatment means with a experimental error as low as possible. In plant breeding such a situation can be exemplified by yield trials comprising a fixed set of varieties and/or hybrids in the final phase of a breeding program. On the other hand, at an intermediate phase when the yield trials include random samples of genetic materials, such as progenies in recurrent selection or topcrosses of inbred lines in hybrid breeding programs, the properties of the population and the selection method must be considered in determining experimental techniques in which plot size is an important factor.

The expected gain from selection (gs%) was taken for plot size determination (Table 3). The gs% values for different proportions of selected progenies and varying plot sizes for ED and GW were obtained from four replications. The variation in gs% with increasing plot size for a fixed proportion of selected progenies depends only on the parameter g% (Table 2) and showed an increasing and asymptotic trend. The expected gain showed a nearly linear increase by decreasing the proportions of selected progenies for a given plot size (Table 3).

The number of progenies to be used for recombination in a recurrent selection program is dictated by the appropriate effective size of the population. For half-sib progenies the effective size is approximately $n = 4N$ (Vencovsky 1987); for example $n = 100$ after recombination of $n = 25$ half-sib progenies. By fixing the number of selected progenies, its proportion will depend on the number of evaluated progenies, and the increase in the expected gain by increasing selection pressure can only be done by enlarging the sample of progenies evaluated. Under similar experimental techniques increases in the number of progenies increases experimental resources that ultimately establishes the limit for the number of progenies.

Under the assumption that the cost of the experimental is proportional to the experimental area, different combinations of p (selection intensity) and x (plot size) can be obtained for a constant area. The underlined values in Table 3 represent some such combinations, including the most commonly used selection intensities (5 to 25%) for family selection. For that particular situation the maximum gain for ED is for plots 3 m² in size. For GW the maximum gain is obtained for plots between 3 and 4 m² and selection intensity between 15 to 20%; for a higher selection pressure the gain will obviously increase and the effective size decrease. For experimental conditions similar to those considered, optimum plot size for the evaluation of half-sib progenies seems to be between 3 to 4 m², or 15–20 plants. The reduction in plot size is always followed by an increase in the sample of progenies under evaluation so that the effective size can be kept nearly constant. The plot sizes agree with those determined by the point of maximum curvature of the CV function for grain yield.

Plot size and number of replications

From the analysis of variance for half-sib progenies, the gain from selection on a progeny mean basis was estimated by $gs = i\,\sigma_p^2(\sigma_p^2 + \sigma^2/r)^{-1/2}$ with truncation selection for both sexes. In percent of the mean it can be shown

Table 3. Estimates of gain from selection in percent of the mean (gs%) for diameter and grain yield for different selection intensities (p) and plot sizes (x)

p(%)	i[a]	Ear diameter[b]					Grain yield[c]				
		(1)	(2)	(3)	(4)	(5)	(1)	(2)	(3)	(4)	(5)
40	0.9659	1.55	1.93	2.21	2.42	2.60	2.45	3.15	3.65	4.06	4.40
35	1.0583	1.69	2.12	2.42	2.65	2.85	2.69	3.46	4.00	4.44	4.82
30	1.1590	1.85	2.32	2.65	2.91	3.13	2.94	3.79	4.39	4.87	5.28
25	1.2711	2.03	2.55	2.90	3.19	$\underline{3.43}$	3.23	4.15	4.81	5.34	$\underline{5.79}$
20	1.3998	2.24	2.80	3.20	$\underline{3.51}$	3.77	3.56	4.57	5.30	$\underline{5.88}$	6.38
15	1.5544	2.49	3.11	$\underline{3.65}$	3.90	4.19	3.95	5.08	$\underline{5.88}$	6.53	7.08
10	1.7550	2.81	$\underline{3.52}$	4.01	4.40	4.73	4.46	$\underline{5.73}$	6.64	7.37	7.99
5	2.0627	$\underline{3.30}$	4.13	4.71	5.17	5.56	$\underline{5.24}$	6.74	7.80	8.66	9.39
2	2.4209	3.87	4.85	5.53	6.07	6.53	6.15	7.91	9.16	10.17	11.03
1	2.6652	4.26	5.34	6.09	6.68	7.19	6.77	8.71	10.08	11.19	12.14

[a] Differential of selection in standard units, from Hallauer and Miranda-Filho (1981)

[b,c] Values estimated from the equations $gs = i\ (1.60)\ x^{0.9249}$ and $gs = i\ (2.54)\ x^{0.9628}$, respectively

that the gain from selection is

$$gs\% = i\,(CV_g)^2\,[(CV_g)^2 + (a^2/r)\,x^{2b}]^{-1/2}, \qquad (1)$$

where CV_g is the genetic coefficient of variation, r is the number of replications, x is the plot size, and *a* and *b* are the parameters of the relation function between CV and plot size. In the experimental evaluation, the total area for each progeny is usually limited by the number of seeds. The total area for one progeny is $S = rx$, and thus $r = S/x$. Replacing r in (1) it follows that:

$$gs\% = i\,CV_g^2\,[CV_g^2 + (a^2/S)\,x^{(2b+1)}]^{-1/2}. \qquad (2)$$

Since b varies between -0.5 and 0, the power of x will be in the limit $1 \geq 2b+1 \geq 0$. Because $2b+1$ is always positive the gain from selection will be greater for smaller plots and, consequently, greater number of replications. Therefore, the best combination of plot size and number of replications for a limited area for each progeny will be given by the minimum plot size, as determined by experimental convenience, followed by maximization of the number of replications. These results agree with those reported by Hatheway (1961), Le Clerg et al. (1962), Thomas and Abou-El-Fithouh (1968), and Storck and Uitdewilligen (1980). The minimum limit for plot size is one plant, but several plants must be taken to represent a plot because: (a) in selection experiments, the evaluation of genotypes is usually on a plot basis, where a sample of plants represents a progeny or genotype; and (b) an estimate of phenotypic variability within plots may be of interest when the within-progeny selection is considered (Miranda-Filho 1987). From (1) it can be seen that the product $i \cdot CV_g$ is the theoretical limit of gain from selection per cycle in percent of the mean; that limit is approximated for high values of r and x that makes $(a^2/r)\,x^{2b}$ tend to zero.

Table 4. Plot sizes (m^2) corresponding to points of maximum curvature of the function $\sigma_m = (a/\sqrt{r})\,x^b$ for varying number of replications (r)[a] and several traits

Trait	1	2	4	6	8	10
Ear number	3.15	2.45	1.90	1.64	1.48	1.36
Ear diameter	1.84	1.45	1.14	0.99	0.90	0.83
Ear yield	3.57	2.75	2.12	1.82	1.64	1.51
Grain yield	3.64	2.81	2.17	1.86	1.67	1.54

[a] r: 1, 2, . . . , 10

The use of the maximum curvature of the CV function for plot size determination has the restriction that it does not consider the number of replications. Such a restriction does not exist if one uses the standard error of the mean (in percentage) instead of the coefficient of variation. The new function is $\sigma_m^2(\%) = (a/\sqrt{r})\,x^b$, where a and b have already been defined. The values of x in the points of maximum curvature of the σ_m function show that for a higher number of replications, smaller plots can be used (Table 4). In Figs. 1 and 2 the values found for the CV function are those shown in Table 4 for the $\sigma_m(\%)$ function with one replication.

The functions relating CV or $\sigma_m(\%)$ with plot size show an asymptotic pattern of variation, and higher levels of precision are attained for plots larger than those corresponding to the points of maximum curvature; thus, such points can be more conveniently considered as minimum size instead of optimum plot size for a given number of replications.

Practical comments

The minimum plot size recommended for use in yield trials depends not only on statistic factors, but also on the practical feasibility of conducting experiments and collecting experimental data. The planting and execution of experiments with 1-m^2 plots followed conventional procedures for yield trials in maize breeding. At this phase, the following points deserve consideration: (a) in the field layout special care must be taken with respect to gross errors such as spacing among and within plots, because small variations of this order may result proportionally in important sources of experimental error; (b) it would be convenient to use border plants in the plot ends because the partial competition of the most extreme plants of micro-plots will cause a non-neglible bias in estimating plot means; (c) a good experimental condition must be assured to keep the final stand as close as possible to the ideal number, because, for example, each blank hill represents a 20% failure in a plot with 5 plants. Planting in excess and thinning at the appropriate phase (Hallauer and Sears 1969) may overcome such a difficulty.

Experimental data on all of the traits that are considered after harvest should be measured in the laboratory. Such a procedure has some advantages because (a) the harvested ears can be then stored to stabilize moisture at a uniform level before data are taken, thus eliminating measuring the moisture levels of the grain for each plot; (b) weighing in the laboratory usually results in more accurate and precise measures, either by using more precise and better calibrated instruments or by avoiding gross mistakes caused by continuous field work; (c) the ear traits, such as damage caused by insects or diseases, genetic defects, ear shape, and kernel type, can be more carefully evaluated.

The comments considered so far are valid regarding the use of small of micro-plots (as small as 1 m^2 or 5 plants) in maize breeding without serious or limiting restrictions. The use of micro-plots is recommended in the following situations: (a) a limited number of seeds are available for each treatment; and (b) the number of entries is too large, as in the first phases of breeding programs. In both instances, the number of replications must be as high as permitted by area limitations. Hallauer (1964) suggested the use of two locations with two replications per location in 2 years for the evaluation of a large number of single, three-way, and double crosses that originated from 60 inbred lines. An increase in the number of locations or replications would result in a small increase in precision for the comparisons of treatment means, but the increase in the experiment would be proportionally greater.

The results presented refer to half-sib progenies, but they could be extended to other kinds of genetic materials, such as top-crosses of inbred lines. A sample of half-sib progenies represents a sample of top-crosses of randomly chosen single-crosses by using the parental population as the tester. In all instances, however, adequate plot techniques are affected by the genetic properties of the base population.

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